High-throughput epitope identification for snakebite antivenom

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High-throughput epitope identification for snakebite antivenom

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Introduction

Insight into the epitopic recognition pattern for polyclonal antivenoms is a strong tool for accurate prediction of antivenom cross-reactivity and provides a basis for design of novel antivenoms. In this work, a high-throughput approach was applied to characterize linear epitopes in 966 individual toxins from pit vipers (Crotalidae) using the ICP Crotalidae antivenom. Due to an abundance of snake venom metalloproteinases and phospholipase A₂s in the venoms used for production of the investigated antivenom, this study focuses on these toxin families.

Objectives

- Identify epitopes in toxins used in immunization
- Characterize tolerated amino acid substitutions in identified epitopes
- Predict cross-reactivity of antivenom

Immunization mixture

To identify epitopes in toxins, peptide microarray experiments were performed. The epitope core sequences are highlighted in blue except for the high-signal epitope in Bothrops asper venom used in antivenom production. The epitope core sequences were localized to surface regions of the toxins.

CLUSTAL O(1.2.1) multiple sequence alignment

The peptide microarrays were used for in silico generation of peptide libraries and experimental setup. The peptide microarrays were used for in silico generation of peptide libraries and experimental setup. The peptide microarrays were used for in silico generation of peptide libraries and experimental setup.

Effect on cross-recognition

The mean AU overlapping peptides for each epitope is shown. The mean AU overlapping peptides for each epitope is shown. The mean AU overlapping peptides for each epitope is shown.

Conclusions

- Custom-designed high density peptide microarray technology enables parallel automated identification of epitopes in hundreds of toxins.
- Integrating multiple sequence alignment allows investigation of the effect of epitope variation on antivenom recognition.
- Cross-reactivity of antivenom is correlated to the degree of conservation in toxin epitopes and flanking residues.